

Patent claims

1. An isolated polynucleotide from coryneform bacteria, comprising a polynucleotide sequence which codes for the lysR2 gene, chosen from the group consisting of
- 5 a) polynucleotide which is identical to the extent of at least 70% to a polynucleotide which codes for a polypeptide which comprises the amino acid sequence of SEQ ID No. 2,
- 10 b) polynucleotide which codes for a polypeptide which comprises an amino acid sequence which is identical to the extent of at least to [sic] 70% to the amino acid sequence of SEQ ID No. 2,
- c) polynucleotide which is complementary to the polynucleotides of a) or b), and
- 15 d) polynucleotide comprising at least 15 successive nucleotides of the polynucleotide sequence of a), b), or c),
- the polypeptide preferably having the activity of the transcription regulator LysR2.
- 20 2. A polynucleotide as claimed in claim 1, wherein the polynucleotide is a preferably recombinant DNA which is capable of replication in coryneform bacteria.
3. A polynucleotide as claimed in claim 1,
- 25 wherein the polynucleotide is an RNA.
4. A DNA as claimed in claim 2 which is capable of replication, comprising
- (i) the nucleotide sequence shown in SEQ ID No. 1, or

T0427060692850

- (ii) at least one sequence which corresponds to sequence (i) within the range of the degeneration of the genetic code, or
- (iii) at least one sequence which hybridizes with the sequences complementary to sequences (i) or (ii), and optionally
- (iv) (iv)[sic] sense mutations of neutral function in (i) which do not modify the activity of the protein/polypeptide.
- 5
- 10 5. A polynucleotide as claimed in claim 2, comprising the nucleic acid sequence as shown in SEQ ID No. 1.
6. The vector pCR2.1lysR2int, which
- 6.1 carries an internal fragment of the citA gene 439 bp in size,
- 15 6.2 the restriction map of which is reproduced in figure 1, and
- 6.3 which is deposited in the E.coli strain TOP10F/pCR2.1lysR2int under no. DSM 13617 at the Deutsche Sammlung für Mikroorganismen und Zellenkulturen [German Collection of Microorganisms and Cell Cultures].
- 20
7. A coryneform bacterium in which the lysR2 gene is attenuated, preferably eliminated, in particular by deletion.
- 25 8. A process for the preparation of L-amino acids, in particular L-lysine and L-valine, which comprises carrying out the following steps,

09826909.072404
T04270.60692860

- a) fermentation of the bacteria which produce the desired L-amino acid and in which at least the lysR2 gene is attenuated,
- 5 b) concentration of the desired product in the medium or in the cells of the bacteria and
- c) isolation of the L-amino acid.
9. A process as claimed in claim 8, wherein
- 10 bacteria in which further genes of the biosynthesis pathway of the desired L-amino acid are additionally enhanced are employed.
10. A process as claimed in claim 8, wherein
- 15 bacteria in which the metabolic pathways which reduce the formation of the desired L-amino acid are at least partly eliminated are employed.
11. A process as claimed in claim 8, wherein
- 20 expression of the polynucleotide(s) which codes (code) for the lysR2 gene is decreased, in particular eliminated.
12. A process as claimed in claim 8, wherein
- 25 the regulatory properties of the polypeptide for which the polynucleotide lysR2 codes are decreased.
13. A process as claimed in claim 8, wherein
- 30 for the preparation of L-amino acids, in particular L-lysine, bacteria in which at the same time one or more of the genes chosen from the group consisting of

09026909.072401

- 13.1 the dapA gene which codes for dihydrodipicolinate synthase,
- 13.2 the eno gene which codes for enolase,
- 13.3 the zwf gene which codes for the zwf gene product,
- 13.4 the pyc gene which codes for pyruvate carboxylase,
- 13.5 the lysE gene which codes for lysine export,
- 13.6 the lysC gene which codes for a feed-back resistant aspartate kinase
- 13.7 the zwal gene which codes for the Zwal protein is or are enhanced, preferably over-expressed, are fermented.
14. A process as claim in claim 8, wherein at the same time one or more of the genes chosen from the group consisting of:
- 14.1 the pck gene which codes for phosphoenol pyruvate carboxykinase,
- 14.2 the pgi gene which codes for glucose 6-phosphate isomerase,
- 14.3 the poxB gene which codes for pyruvate oxidase,
- 14.4 the zwa2 gene which codes for the Zwa2 protein,
- 14.5 the hom gene which codes for homoserine dehydrogenase
- 14.6 the thrB gene which codes for homoserine kinase, and

14.7 the panD gene which codes for aspartate decarboxylase

is or are attenuated, in particular eliminated.

15. A process as claimed in claim 8,
5 wherein
for the preparation of L-amino acids, in particular L-
valine, bacteria in which at the same time one or more
of the genes chosen from the group consisting of
15.1 the ilvBN gene which codes for acetohydroxy-acid
10 synthase,
15.2 the ilvD gene which codes for dihydroxy-acid
dehydratase,
15.3 the mqo gene which codes for malate:quinone
oxidoreductase
15 is or are enhanced, in particular over-expressed, are
fermented.
16. A process as claimed in one or more of the preceding
claims,
wherein
20 microorganisms of the species *Corynebacterium*
glutamicum or *Brevibacterium lactofermentum* are
employed.
17. A process for discovering RNA, cDNA and DNA in order
to isolate nucleic acids, or polynucleotides or genes
25 which code for the transcription regulator LysR2 or
have a high similarity with the sequence of the lysR2
gene,
which comprises
employing the polynucleotide sequences as claimed in
30 claims 1 to 4 as hybridization probes.

18. A process as claimed in claim 15,
wherein arrays, micro arrays or DNA chips are
employed.

09826909 072401
T04270 60692860